MICROBIC VIRULENCE AND HOST SUSCEPTIBILITY IN PARATYPHOID-ENTERITIDIS INFECTION OF WHITE MICE.

VIII. THE EFFECT OF SELECTIVE BREEDING ON HOST RESISTANCE. FURTHER STUDIES.

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In a previous paper we reported a series of experiments relating to the effect of selective breeding on the resistance of mice to mouse typhoid infection and mercury bichloride poisoning. This work has now progressed to a point where further studies may be described, and a general summary stated.

All technique has been similar to that employed before. Mouse typhoid bacilli, the epidemic M. T. II strain of Bacillus pestis caviae, were administered to mice intrastomachally in known quantities, by means of a silver catheter and syringe. The Rockefeller Institute breed of mice, whose history and maintenance have been described, was used throughout.

The published experiments relating to this study may be reviewed as follows: At the outset, 100 Rockefeller Institute mice were inoculated per os with a given dose of mouse typhoid bacilli. 2 months later, ten survivors, males and females, which showed no mouse typhoid bacilli in blood or feces, and no specific agglutinins, were bred in the stock mouse room. Later, when fourteen of the resulting F1 generation had reached a weight of 16 to 18 gm. each, they were inoculated in the same manner, together with twenty unselected mice of the same original strain as controls. 28 per cent of these survived, and 48 per cent of the F1 selected mice. Subsequently, six of the F1 survivors, free of mouse typhoid bacilli, were bred as before and 3 months later, twenty-one F2 offspring and twenty-one controls were inoculated. 30 per cent of the controls and 85 per cent of the F2 mice survived. This repeated selection and breeding of the resistant individuals of each generation

had apparently given rise to a strain of mice considerably more refractory to in-
fection than the parent stock.

Since the publication of the above experiments, this procedure has 
been continued for three more generations.

Experiment I.—Ten survivors of the F₂ generation, free of mouse typhoid bacilli 
in blood and stools, were bred in the mouse stock room. On May 9, 1924, twenty-
one of the F₃ offspring, together with a control group of 50 mice, were injected 
per os with the routine dose of mouse typhoid bacilli. Dead mice were autopsied 
and cultured as in the previous work.

The mortality of the F₃ generation and of the Rockefeller Institute 
controls is recorded in Text-fig. 1. The death rate and final mortality 

![Text-fig. 1. The susceptibility of the “resistant strain,” fourth generation, 
to mouse typhoid infection.]

of the selected mice were again much lower than those of the control 
group. But in addition to this, another fact was brought out by the 
experiment; namely, that influences which modify, in a seasonal man-
ner, the susceptibility of different races of mice to mouse typhoid 
infection, affect this group also. For, just as the mortality of five 
other races of mice injected at this time was higher than at any other 
period during the year, so the resistant group succumbed in larger 
numbers than before.

Only three survivors of the F₃ group were free of mouse typhoid bacilli in feces 
and specific blood agglutinins. These, two females and one male, were mated in 
the breeding room. 12 weeks later, fifteen F₄ offspring were available, but instead

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4 Pritchett (see Text-figs. 1 and 2).
of being inoculated as before, they were mated, brother to sister. On February 5, 1925, 42 of the sixth generation, F6, together with a routine monthly series of 50 Rockefeller Institute controls, 50 Bagg, 50 Lathrop, and 50 Little mice (dilute brown), and also a group of so called "susceptible mice," which will be described later, were inoculated with the routine dose of mouse typhoid culture.

At this time, unfortunately, it was noticed that many of the F6 mice were puny, underweight, cold, and sickly. No explanation was at hand, so the group was used. That some disturbing element had appeared was evidenced by the mortality rate thereafter. The unhealthy mice succumbed quickly, and in numbers sufficient to bring the total mortality slightly higher than that of the unselected controls (Text-fig. 2). The mortality was more gradual, however, and would have been quite as usual if the weaklings dying from the 7th to the 10th days had been discarded.
While this series of experiments on the selective breeding of resistant mice was in progress, another strain was developed from the same original Rockefeller Institute stock by the repeated selection and breeding of the most susceptible individuals from a random group.

Experiment 2.—On March 5, 1924, twenty-one female mice, with litters 10 to 12 weeks old, were taken at random from the Rockefeller Institute stock, and numbered consecutively. The young mice were left in their respective cages, while the mothers were each placed in a separate jar with marks of identification, and taken to the experimental room. Each then received per os the usual dose of about 4,000,000 mouse typhoid bacilli in ½ cc. volume of broth.

The mortality of this group is shown in Text-fig. 3. The curve corresponds closely to the standard control curve for this race of mice. Deaths were noted on the 5th day and continued quite regularly until the 18th day, after which only one fatality was recorded.

The litters of the most susceptible mothers, that is the first six mice to die, were selected, and twenty of these females were mated with four of the males. The F₂ offspring were old enough to be weaned on May 22, 1924. The cages and F₁ mothers were then numbered, and the latter were placed in separate jars and injected as before, together with a random group of twenty Rockefeller Institute controls.

Both series reacted quite similarly to the mouse typhoid bacilli (Text-fig. 4). The mortality of the selected mice was slightly

\[ \text{Webster, L. T., J. Exp. Med., 1923, xxxvii, 251.} \]
greater than that of the control group, but the differences could not be considered especially significant.

A second selection was made. The female offspring of the first six \( F_1 \) mothers to die were mated with four males from the first two of these litters. On August 15 the \( F_2 \) offspring were weaned and the twenty \( F_2 \) mothers were numbered accordingly, and taken to the experimental room for inoculation. All conditions and injection technique were kept uniform. 50 unselected Rockefeller Institute mice were inoculated as controls at the same time.

Text-fig. 5 compares the mortality of the two series. After the 8th day deaths were recorded frequently in both groups. On the 16th day 48 per cent of the controls and 65 per cent of the susceptibles were dead, and after 2 months the final mortality of the controls was 62 per cent, as compared to 80 per cent of the \( F_2 \) selected mice.

For a third time the litters of the most susceptible mothers were inbred and, on November 14, when the offspring had been weaned, the \( F_3 \) mothers, together with 50 Rockefeller Institute controls, and 50 of each of three other races, were given the usual dose of mouse typhoid bacilli.

The death rate of the selected mice was high and the final mortality 85 per cent (Text-fig. 6). The mortality of the control group (48 per cent), as well as that of the Bagg, Lathrop, and Little mice (dilute brown) strains, was very low.\(^6\)

\(^6\) Further details relating to the control curves in this series will be published by Dr. I. W. Fritchett.
A selection of offspring of the most susceptible individuals of the F3 group was again made, and another breeding experiment carried out. On February 14, 1925, fourteen F4 mothers were taken to the experimental room and, together with the routine monthly series of 150 mice, and the "resistant group" described above, were inoculated with the routine dose of mouse typhoid bacilli.

The mortality rates and figures of the "resistant," "susceptible," and "control" groups are plotted in Text-fig. 2, from which it may be seen that the susceptible mice died more rapidly and in larger numbers than either of the other series.

DISCUSSION.

The natural resistance of mice to pathogenic bacteria has been a subject of our studies for some time. In the first place, we have found that this quality of resistance is present in different amounts in individuals of the same family or race, and that these differences, under properly controlled conditions, take the form of a frequency curve. Furthermore, by an artificial selection of especially resistant or susceptible individuals it has been possible to breed strains at will, whose average resistance is greater or less than that of the original random group. It seems probable, then, that successive descendants of two given individuals inherit definite amounts of potential resistance, which vary according to the law of probability about a mean which approximates the mean resistance of the original pair.

It would seem that racial differences in resistance to mouse typhoid infection can be expressed by a relatively constant value.
Finally, whatever the potential inherited constitution of individuals, families, or races may be, it is affected profoundly by seasonal influences, food, and general hygienic conditions. For this reason it is erroneous to speak of the presence or absence of resistance as though it were a unit factor. Rather it should be considered a manifestation of an extremely complex mechanism, modified by heredity and environment, whose quantity in individuals of a large group tends to follow the laws of chance, about a mean which is more or less a characteristic of the race.

CONCLUSIONS.

1. Survivors of an experimental mouse typhoid infection, selected and bred for a number of consecutive generations, give birth to offspring which are more resistant to the disease than a random group of the same inbred race.

2. Offspring of females most susceptible to this infection give birth to individuals which are more susceptible than a group of similar unselected mice.